Status of umbilical cord blood transplantation in the year 2001

J M Hows

Abstract

Umbilical cord blood (UCB) transplantation is limited to small recipients because of the low haemopoietic cell dose. Children from ethnic minority groups may benefit most from cord blood transplantation. Cohort controlled retrospective data indicate that there is significantly less acute and chronic graft versus host disease associated with the transplantation of human major histocompatibility complex (HLA) identical sibling cord blood compared with HLA identical sibling marrow. Controlled data are not yet available to confirm this observation in unrelated donor cord blood transplantation. The difference in leukaemic relapse seen after cord blood compared with bone marrow transplantation is also unknown. Tentative recommendations for the use of umbilical cord blood for transplantation are as follows. Collection is indicated from healthy newborn siblings when urgent transplantation is required for an older child in a family. The haematologist responsible for the older child, with the approval of the family and the obstetric team, should contact the medical director of the nearest cord blood bank to discuss arrangements for the UCB to be collected and HLA typed. Antenatal blood sampling to HLA type the fetus is not recommended. Umbilical cord blood should be considered when allogeneic transplantation is the treatment of choice for a child who does not have an HLA identical sibling, or a well matched unrelated adult volunteer donor. The potential advantages and disadvantages of using an HLA haplo-type matched peripheral blood stem cell family donor rather than an unrelated cord blood donation should be discussed. There are no comparative data available

as yet. At present, UCB transplantation should only be considered if a suitably matched donation contains at least 2×10^7 kg nucleated cells. Effectively, this means that most adults and larger children are not suitable recipients.

(*J Clin Pathol* 2001;54:428–434)

Keywords: umbilical cord blood transplantation; graft versus host disease; bone marrow transplantation; leukaemia; cell expansion

In 1987, a child with Fanconi's anaemia received an allogeneic transplant using the cryopreserved umbilical cord blood (UCB) collected from his human major histocompatibility complex (HLA) identical sibling. The successful transplant took place in Paris with close international collaboration from scientists and clinicians in the USA.¹ As a result of this report, the potential of UCB as a source of haemopoietic stem cells (HSCs) for transplantation rapidly became an area of intense clinical and scientific interest. Several probable or possible advantages of UCB as a source of HSCs have been quoted frequently in the literature, very often without sound clinical or in vitro evidence (table 1). In the year 2001, the characteristics of UCB have been more thoroughly investigated in vitro and the quality of clinical transplant studies has improved. Therefore, it is becoming easier to appreciate the true place of UCB as a source of HSCs. This leader reviews the current issues surrounding UCB transplantation.

Alloreactivity of UCB

Optimistic early reports of UCB transplantation suggested the virtual absence of clinically severe graft versus host disease (GVHD).² In vitro studies of alloreactivity of UCB mononuclear cells (MNCs) produced conflicting results, ranging from greatly reduced³ to the

Table 1 Possible advantages of umbilical cord blood (UCB) compared with bone marrow as a source of unrelated haemopoietic stem cells for transplantation (SCT)

Advantages quoted for UCB	Comment
Rapid stem cell donor selection	Yes if "donor" is a newborn HLA identical sibling. Potentially, searches of unrelated cord blood banks should be rapid. However, controlled clinical data comparing efficiency of UCB banks with volunteer stem cell donor registry searches are not yet available
Negligible transmission of CMV to the recipient	True, because <0.1% of healthy neonates are CMV positive, compared with 10–60% of adult volunteer donors
No donor morbidity	True, as long as UCB is collected without influencing the management of mother or child
Enhances availability of unrelated stem cell donations to non-white patients	Some data to support. UCB donation is more acceptable than volunteer bone marrow donation to some non-white ethnic groups
UCB transplants are associated with less GVHD than SCT from older donors	HLA partially mismatched UCB transplants are less alloreactive than adult donor stem cell transplants (SCT). Retrospective cohort controlled data support this, but only after HLA identical sibling SCT ¹²

CMV, cytomegalovirus; GVHD, graft versus host disease; HLA, human major histocompatibility complex antigens.

Division of Transplantation Sciences, Blue Zone A, Entrance 11, Southmead Health Services, University of Bristol, Westbury on Trym, Bristol BS10 5NB, UK
J M Hows

Correspondence to:
Professor Hows
jill.hows@bristol.ac.uk

Accepted for publication
1 February 2001

same or greater alloreactivity compared with adult peripheral blood mononuclear cells.^{4,5} Consistent findings have been (1) the “naïve” immunophenotype of UCB lymphocytes and,⁶ (2) that alloantigen primed UCB T cells are relatively unresponsive to the original stimulator in secondary mixed lymphocyte reactions.^{7,8}

Graft versus host disease

As clinical experience increased it became apparent that clinically relevant acute GVHD (AGVHD) can occur after UCB transplantation, even when the donation is from an HLA genotypically identical sibling.^{9,10} AGVHD is even more frequent after HLA haplotype mismatched family donor transplants¹⁰ or transplantation using partially matched unrelated donations obtained from the growing number of cord blood banks worldwide.¹¹

Over the first decade of clinical experience, the lack of prospective randomised studies or even retrospective cohort controlled studies made realistic assessment of the comparative probability of acute and chronic GVHD after UCB and marrow stem cell transplants (SCTs) difficult. The first controlled study was recently performed by the International Bone Marrow Transplant Registry and the Eurocord Group.¹² This retrospective cohort controlled analysis compared the outcome of UCB with bone marrow SCTs in children under the age of 15 years transplanted for malignant and non-malignant haematological conditions. Transplants were carried out between 1990 and 1997, with 2018 children receiving unmanipulated bone marrow and 106 UCB stem cells. There were significant differences in patient and protocol characteristics between the two cohorts. Statistical corrections were made to account for these in the final analysis. Importantly, the bone marrow transplant group were older ($p < 0.0001$), more likely to have acute leukaemia ($p < 0.0001$), had a shorter diagnosis to transplant interval ($p = 0.0001$), were more likely to receive methotrexate in addition to cyclosporin as post transplant immunosuppression ($p < 0.0001$), and were less likely to receive prophylactic recombinant haemopoietic growth factors in the post transplant period ($p < 0.0001$). The median nucleated cell dose received by the UCB stem cell recipients was almost one log lower than that received by the bone marrow stem cell recipients: $0.47 \times 10^6/\text{kg}$ compared with $3.5 \times 10^6/\text{kg}$, respectively. The probability of survival at three years was not significantly different between the two cohorts. The causes of death between the two cohorts were similar and unremarkable, although follow up was too short to assess the impact of graft source on the probability of recurrent disease and death as a result of relapse. Thus, it remains uncertain whether allogeneic UCB has the same graft versus leukaemia potential as bone marrow. Multifactorial analysis clearly indicated that UCB was associated with significantly less acute ($p = 0.001$) and chronic ($p = 0.02$) GVHD than bone marrow. Whether these observations are the result of reduced alloreactivity of UCB

lymphocytes or the five to 10 times lower numbers of lymphocytes transplanted compared with unmanipulated bone marrow is unknown. It remains to be seen whether the same observations hold true for children transplanted with HLA “matched” unrelated and HLA haplotype mismatched family donations.

Engraftment potential of UCB haemopoietic cells in preclinical studies

In vitro studies have emphasised the proliferative superiority of primitive UCB haemopoietic cells compared with adult bone marrow. Mixed colony forming cells (CFC mix) and longterm culture initiating cells (LTC-ICs) in UCB demonstrated more proliferative potential than those in bone marrow.^{13,14} Wang *et al* have shown a significantly higher frequency of human “SCID (severe combined immunodeficiency) repopulating cells” (SRCs) in UCB than resting bone marrow in a xenogeneic immunodeficient mouse model.¹⁵ It is generally accepted that the human SRC is a more primitive cell than most LTC-ICs, predominantly having the $\text{CD}34^+38^-$ phenotype and being capable of multilineage repopulation in non-obese diabetic (NOD)/SCID recipients.¹⁶ It is probable that a minority of the most primitive SRCs have the $\text{CD}34^-$ phenotype.¹⁷

Engraftment potential of UCB HSCs in clinical transplantation

In parallel with the preclinical studies described above, the high proliferative capacity of bone marrow repopulating cells in UCB has been confirmed in clinical transplants. Gluckman and the Eurocord Group first emphasised that around 3×10^7 nucleated cells/kg of recipient body weight was sufficient for primary and sustained engraftment.¹⁰ Additional clinical studies confirmed that reliable engraftment occurs with $0.3\text{--}0.4 \times 10^6/\text{kg}$ $\text{CD}34^+$ cells after UCB transplantation,¹⁸ suggesting that UCB HSCs have high proliferative potential in clinical transplantation. A substantial body of clinical data now exists confirming that approximately one tenth of the nucleated and $\text{CD}34^+$ cell dose routinely given in bone marrow grafts is associated with successful engraftment after UCB transplantation.^{18–20} Despite these encouraging data, the limited haemopoietic cell dose in individual UCB donations has emerged as the most important disadvantage of UCB as a source of HSCs for clinical transplantation.

Several investigators have observed that the median time to neutrophil and platelet recovery after UCB transplantation is longer than that expected after marrow or adult peripheral blood stem cell (PBSC) transplants.^{10,18,19} The delay in platelet recovery is more pronounced than that of neutrophils, partly because neutrophil recovery is often enhanced by the use of post transplant recombinant haemopoietic growth factors *in vivo*.²¹ Figure 1 shows how the infused $\text{CD}34^+$ cell dose has a large impact on initial peripheral blood platelet (fig 1A) and neutrophil (fig 1B) recovery after unrelated UCB transplantation (J Wagner and J Kurtzberg, 1999, reproduced with permission). The

Figure 1 The relation between the infused CD34⁺ cell dose and (A) platelet and (B) neutrophil recovery after unrelated umbilical cord blood transplantation. The numbers within the graphs refer to the dose of CD34⁺ cells ($\times 10^6/\text{kg}$). There is a strong direct correlation between cell dose and peripheral blood recovery. Transplants containing less than $0.3 \times 10^6/\text{kg}$ CD34⁺ cells have a high probability of poor engraftment or non-engraftment.

infused cell dose after unrelated donor transplantation correlated with post transplant survival (fig 2A). Interestingly, the correlation between HLA mismatch and survival (fig 2B) was less impressive than the association with infused cell dose.

Attempts to optimise collection of UCB donations

There has been considerable cord blood banking activity worldwide over the past decade. In some countries there has been massive central financial support, whereas in others, including the UK, all banking activity to date has been financed through research and development funds raised by individual cord blood banks. Up to date information on the content of cord blood banks worldwide can be accessed at the bone marrow donors worldwide web site (<http://www.bmdw.org/>). At the time of writing, over 30 000 cord blood donations are banked and available for patients worldwide, with approximately 4000 in the UK.

Much collaborative work has been done to optimise the haemopoietic cell content of UCB donations for banking.²²⁻²⁴ Studies of the predictive impact of obstetric factors on haemopoietic cell yield have been performed. Direct correlations were shown between total nucleated blood cell count and the volume of blood collected ($p < 0.001$). The total nucleated blood cell count directly correlated with length of gestation ($p < 0.001$) and length of labour ($p = 0.002$). An inverse correlation was shown between time from delivery of the infant to clamping of the umbilical cord ($p = 0.02$). A significantly higher nucleated cell count was obtained in collections from primiparous compared with multiparous deliveries ($p < 0.001$).²⁵ These data may be used to select

Figure 2 The effect of CD34⁺ cell dose on survival after unrelated umbilical cord blood transplantation. (A) There is a direct correlation between increasing cell dose and the probability of survival at one year post transplant. Patients receiving less than $0.3 \times 10^6/\text{kg}$ CD34⁺ cells have a significantly higher probability of death in the first year after transplantation than those receiving a higher CD34⁺ cell dose. The numbers within the graphs refer to the dose of CD34⁺ cells ($\times 10^6/\text{kg}$). (B) The relatively minor effect of human major histocompatibility complex (HLA) mismatch on survival compared with the major effect of cell dose. HLA typing was performed by intermediate resolution DNA methodology. mm, number of HLA loci mismatched.

optimal deliveries for collection, thus making cord blood banking as cost effective as possible. However, it is important that the way the birth is managed is not altered in any way by cord blood donation. The time of clamping the umbilical cord in relation to the birth of the infant should always remain the responsibility of the obstetric team.

In Bristol, the employment of experienced midwives with a dual role as midwife counsellors and cord blood collectors reduced the discard rate of low volume collections greatly (table 2).²² However, despite optimal collection and processing procedures only a small minority of donations in the Bristol cord blood bank contain sufficient cells for recipients over 50 kg in weight. The histogram in fig 3 demonstrates this by indicating the proportion of recipients of different sizes for whom an optimal donation ($> 3.7 \times 10^7/\text{kg}$ nucleated cells) or a just sufficient donation ($> 2.0 \times 10^7/\text{kg}$ nucleated cells) is available in the Bristol bank. With the exception of when the potential recipient has a common HLA type, the chance of finding a large enough donation for an adult with two or less HLA mismatches using high resolution DNA typing is very low. High infused cell dose

Table 2 Optimisation of cord blood donations in the Bristol cord blood bank

Number of donations	749
Mean volume collected (SD)	98 ml (32)
Mean total nucleated blood cell count (SD)	10.2×10^8 (4.6)
Mean total CD34 ⁺ count (SD)	4.3×10^6 (3.8)
Donations discarded because of small volume (<60 ml)	56 (7.5%)

Figure 3 The proportion of recipients of different weight for whom an optimal cord blood donation ($> 3.7 \times 10^7$ /kg nucleated cells) or a just sufficient donation ($> 2.0 \times 10^7$ /kg) of nucleated blood cells is available in the Bristol cord blood bank. With the exception of when the potential recipient has a common HLA type, the chance of finding a large enough donation for an adult with two or less human major histocompatibility complex (HLA) mismatches using high resolution DNA typing is very low.

is an important predictor for post transplant survival, independent of the stem cell source.²⁶ It is clear that additional research is required before UCB can be considered to be a safe source of stem cells for larger children and adults.

In vitro expansion using haemopoietic growth factors

Our own expansion experiments were designed to facilitate easy and economical scaling up for future clinical use.²⁷ A 50 fold expansion of colony forming cells (CFCs) and CD34⁺ cells and a three to sixfold expansion of LTC-ICs was observed. We also calculated that, using our current in vitro expansion protocol and allowing for cell losses during processing, the expansion of 20% of an average donation infused with the remaining 80% of the unprocessed donation would provide an overall five to sevenfold increase in CFCs and CD34⁺ cells

Table 3 In vitro expansion of 20% of an umbilical cord blood donation

	CD34 ⁺	CFU-GM	LTC-IC
Fraction used for expansion	0.2 unit*	0.2 unit	0.2 unit
Post processing fraction	0.15 unit**	0.15 unit	0.15 unit
Fold expansion	×43	×50	×3
Post expansion equivalent	6.5 units	7.5 units	0.45 units
Total yield (unit equivalent)	6.5 + 0.8† = 7.3	7.5 + 0.8 = 8.3	0.45 + 0.8 = 1.25

*One unit represents 20% cell content of one umbilical cord blood donation.

**Unit equivalent allowing for cell losses during processing.

†0.8 represents 0.8 units or 80% of an umbilical cord blood unit infused without in vitro expansion.

CFU-GM, granulocyte-macrophage colony forming units; LTC-IC longterm culture initiating cells.

Table 4 In vitro expansion of long term culture initiating cells (LTC-IC) in serum replete and serum depleted culture conditions in four paired experiments

Experiment	1	2	3	4
LTC-IC freq on day 0*	1/26	1/22	1/24	1/55
Total cells on day 0	1 × 10 ⁵	1 × 10 ⁵	1 × 10 ⁵	1 × 10 ⁵
Total LTC-IC on day 0	3846	4545	4167	1818
<i>Serum replete</i>				
LTC-IC freq on day 14	1/14 285	1/14 706	1/10 753	1/20 000
Total cells on day 14	2.24 × 10 ⁸	2.91 × 10 ⁸	2.18 × 10 ⁸	2.23 × 10 ⁸
Total LTC-IC on day 14	15 681	19 787	20 273	11 150
Fold expansion	4.1	4.4	4.9	6.1
<i>Serum depleted</i>				
LTC-IC freq on day 14	1/10 417	1/3957	1/2710	1/3906
Total cells on day 14	1.49 × 10 ⁸	0.76 × 10 ⁸	0.47 × 10 ⁸	0.39 × 10 ⁸
Total LTC-IC on day 14	14 304	19 202	17 269	9959
Fold expansion	3.7	4.2	4.1	5.5

Day refers to the number of days of expansion culture.

Freq, frequency.

and maintenance of LTC-ICs (table 3). In serum free conditions suitable for clinical studies, the CD34⁺ population was expanded more, with less expansion of mature CD34⁺ cells than in serum replete conditions.²⁸ Longterm culture initiating cell expansion was similar in serum replete and serum depleted conditions (table 4).

The effect of infusing 20–40% of a clinical donation expanded in vitro on clinical engraftment is difficult to calculate. Preliminary clinical studies indicate that when 20% of the donation is in vitro expanded and 80% given unmanipulated the transplant is well tolerated, but no significant change in the time to neutrophil or platelet recovery is observed compared with historical controls.^{29,30} These clinical observations do not provide additional information on the fate of expanded cells in vivo. Therefore, we have investigated the effect of in vitro expansion on the primitive multipotent human SCID repopulating cell in the NOD/SCID mouse before embarking on clinical studies.

Characteristics of human SRCs in the NOD/SCID mouse model

An assay for primitive human haemopoietic cells based on their ability to repopulate the bone marrow of SCID mice after intravenous injection has been developed. Transplantation of human bone marrow or UCB results in engraftment of primitive cells that proliferate and show multilineage differentiation into LTC-ICs, CFCs, mature myeloid cells, and B cells.^{31,32} The engrafting cell has been termed the SCID repopulating cell (SRC). Most SRCs have the primitive CD34⁺ CD38⁺ phenotype.³³ NOD/SCID mice provide a higher and more consistent level of human engraftment than do SCID mice because they have defective natural killer (NK) cells, in addition to absent B and T cell function.³⁴ Limiting dilution analysis has demonstrated the frequency of SRCs in UCB and bone marrow nucleated cells to be 1 in 9.3×10^5 and 1 in 3.0×10^6 , respectively.¹⁵ In our own laboratory, the SRC frequency in cord blood has been calculated as 1 in 1.5×10^4 CD34⁺ cells.³⁵

We have carried out a series of experiments to test the effect of in vitro expansion of cord blood CD34⁺ cells on SRC frequency. Based on previous work in our laboratory, we injected MNCs containing 3×10^4 CD34⁺ cells, or 1–2 SRCs.³⁵ We hypothesised that if there was significant loss of SRCs as a result of in vitro expansion, mice receiving 3×10^4 expanded CD34⁺ cells would fail to engraft.³⁶ Cord blood CD34⁺ cells were cultured in vitro for three, seven, or 10 days in stem cell factor (SCF), Flt3 ligand (Flt3), thrombopoietin (TPO), interleukin 3 (IL-3), IL-6, and granulocyte colony stimulating factor (G-CSF), all at 10 ng/ml. Seventy six mice received fresh or cultured cells from eight different cord bloods. Figure 4A shows that SRCs are still present after three, seven, and 10 days of culture. The mean level of engraftment with cultured cells was lower (1%) compared with fresh cells (7.4%) when the same number of expanded or fresh CD34⁺

Figure 4 Engraftment of human cord blood cells with or without *in vitro* expansion in non-obese diabetic/severe combined immunodeficient (NOD/SCID) mice after five weeks. Cord blood CD34⁺ cells were cultured *in vitro* for three, seven, or 10 days in stem cell factor (SCF), Flt3 ligand (Flt3), thrombopoietin (TPO), interleukin 3 (IL-3), IL-6, and granulocyte colony stimulating factor (G-CSF), all at 10 ng/ml. Seventy six mice received fresh or cultured cells from eight different cord bloods. (A) SRC are still present after three, seven, and 10 days of culture. The mean level of engraftment with cultured cells was lower than with fresh cells (1% v 7.4%) when the same number of expanded or fresh CD34⁺ cells was transplanted. This suggests that although engraftment of expanded cells occurs there is less efficient proliferation and differentiation *in vivo*, resulting in a lower proportion of human cells in the bone marrow after five weeks. (B) If the input number of expanded cells is reduced, engraftment is seen with the expanded progeny of only 0.6×10^4 CD34⁺ cells. This suggests that some expansion of SRC numbers may occur *in vitro*.

cells was transplanted. This suggests that although engraftment of expanded cells occurs, there is less efficient proliferation and differentiation *in vivo*, resulting in a lower proportion of human cells in the bone marrow after five weeks. Figure 4B shows that if the input number of expanded cells is reduced, engraftment is seen with the expanded progeny of only 0.6×10^4 CD34⁺ cells. This suggests that some expansion of SRC numbers may occur *in vitro*.

In conclusion, *in vitro* culture at least maintains SRC numbers, with a possible two to fivefold expansion. Despite this, there is some evidence to suggest that the proliferative capacity of SRCs is reduced by *in vitro* expansion. *In vitro* expansion may limit the proliferative capacity of SRCs *in vivo*. In addition, cytokine exposure *in vitro* may reduce the efficiency of specific migration and homing of SRCs to the bone marrow.³⁷

At this stage, it is not safe to extrapolate the preclinical findings using the NOD/SCID mouse model to clinical cord blood transplantation in general. Thus, expansion of cord blood cells in the clinical setting should only be undertaken in highly specialised centres with appropriate laboratory backup. Expansion should not be attempted in patients in whom a backup source of autologous or allogeneic stem cells is not available. The impact of *in vitro* expansion of part of a UCB donation on engraftment can only be evaluated by gene

marking studies, which at present remains technically challenging.

Investigation of increasing haemopoietic cell dose by transplantation of multiple UCB donations

Investigators have explored the possibility of using multiple donations to enhance early engraftment in larger recipients receiving UCB transplants. There are two preliminary reports in the literature of using multiple UCB donations to increase cell dose in clinical UCB transplantation.^{38,39} In both cases, four to 12 HLA unmatched donations were infused into either paediatric or adult recipients. In Shen's report the patients did not receive myeloablative conditioning and haemopoietic chimaerism was not adequately studied, making interpretation of these data difficult.³⁸ In the report by Weinreb *et al*, myeloablative conditioning was given to a 67 kg adult patient with advanced chronic myeloid leukaemia. Early haemopoietic recovery was satisfactory, with evidence from molecular HLA typing to detect haemopoietic mixed chimaerism that multiple donations had engrafted. Later, one of 12 donations predominated. Of interest, the donation providing longer term engraftment was one HLA haplotype matched with the recipient, the other donations were completely mismatched. The patient relapsed on day 50, there was no acute GVHD.³⁹ Because it is relatively easy to monitor the

engraftment of individual donations by following a molecular marker such as HLA, it should be possible to evaluate multiple donations as a way of increasing haemopoietic cell dose after clinical transplants.

Conclusions

The past 10 years has provided laboratory and clinical data on the validity of UCB as a useful source of haemopoietic stem cells for transplantation. The most exciting clinical finding is that UCB transplantation is associated with less acute and chronic GVHD than bone marrow.¹² It is important to remember that this observation was made in the context of HLA identical sibling paediatric transplantation, and that the probability of disease recurrence is not yet known. These findings should not be extrapolated to unrelated donor transplants or to UCB transplantation in adults. Comparative data on adult and unrelated donor UCB donation are still awaited. However, it is already apparent that altruistic cord blood donation to extend the unrelated stem cell donor pool is popular in non-white ethnic groups. Cord blood transplantation is valuable for children from non-white backgrounds, who remain disadvantaged by the relatively small number of ethnically matched volunteers in marrow donor registries. In the UK, the north London cord blood bank has made an exceptional effort to promote cord blood donation by non-white individuals.⁴⁰ Finally, the success of UCB transplantation must be compared with other emerging transplant strategies used when allogeneic transplantation is the treatment of choice but an HLA identical sibling donor is not available. HLA haploidentical family members donating PBSCs mobilised by recombinant G-CSF are a promising source of alternative donors.⁴¹

Tentative recommendations for the use of UCB for transplantation

UCB collection is indicated from healthy newborn siblings when urgent transplantation is required for an older child in a family. Collection of the cord blood should be done by trained personnel rather than the obstetric staff in charge of the delivery. Limited NHS resources in the UK restrict this service to specialised National Blood Service Centres, these include centres in north London, Newcastle, and Bristol. The haematologist responsible for the older child, with the approval of the family and the obstetric team, should contact the medical director of the nearest cord blood bank to discuss arrangements for the UCB to be collected and HLA typed. Antenatal testing of the baby's HLA type is not recommended.

UCB should be considered when allogeneic transplantation is the treatment of choice for a child who does not have an HLA identical sibling or a well matched unrelated adult volunteer donor. The potential advantages and disadvantages of using an HLA haplotype matched PBSC family donor rather than an unrelated cord blood donation should be discussed. There are no comparative data available as yet.

At present, UCB transplantation should only be considered if a suitably matched donation contains at least $2 \times 10^7/\text{kg}$ nucleated cells. Effectively, this means that most adults and larger children are not suitable recipients.

- 1 Gluckman E, Broxmeyer HA, Auerbach AD, *et al.* Hematopoietic reconstitution in a patient with Fanconi's anemia by means of umbilical-cord blood from an HLA-identical sibling. *N Engl J Med* 1989;321:1174-8.
- 2 Wagner JE, Broxmeyer HE, Byrd RL, *et al.* Transplantation of umbilical cord blood after myeloablative therapy: analysis of engraftment. *Blood* 1992;79:1874-81.
- 3 Harris DT, Schumacher MJ, Locascio J, *et al.* Phenotypic and functional immaturity of human umbilical cord blood T lymphocytes. *Proc Natl Acad Sci U S A* 1992;89:10006-10.
- 4 Haque K, Truman C, Dittmer I, *et al.* Frequencies of cytotoxic T lymphocyte precursor estimate in three different populations. *Asian Pac J Allergy Immunol* 1999;17:93-9.
- 5 Deacock SJ, Schwarer AP, Bridge J, *et al.* Evidence that umbilical cord blood contains a higher frequency of HLA class II-specific alloreactive T cells than adult peripheral blood. A limiting dilution analysis. *Transplantation* 1992;53:1128-34.
- 6 Roncarolo MG, Bigler M, Martino S, *et al.* Immune functions of cord blood cells before and after transplantation. *J Hematother* 1996;5:157-60.
- 7 Risdon G, Gaddy J, Horie M, *et al.* Alloantigen priming induces a state of unresponsiveness in human umbilical cord blood T cells. *Proc Natl Acad Sci U S A* 1995;92:2413-17.
- 8 Hagihara M, Chargui J, Gansuud B, *et al.* Umbilical cord blood T lymphocytes are induced to apoptosis after being allo-primed in vitro. *Bone Marrow Transplant* 1999;24:1229-33.
- 9 Wagner JE, Kernan NA, Steinbuch M, *et al.* Allogeneic sibling umbilical-cord-blood transplantation in children with malignant and non-malignant disease. *Lancet* 1995;346:214-9.
- 10 Gluckman E, Rocha V, Boyer-Chamard A, *et al.* Outcome of cord-blood transplantation from related and unrelated donors. Eurocord transplant group and the European blood and marrow transplantation group. *N Engl J Med* 1997;337:373-81.
- 11 Arcese W, Iori AP, Screnci M, *et al.* Umbilical cord blood transplant from HLA-mismatched unrelated donor in high-risk leukemia. *Bone Marrow Transplant* 1998;21(suppl 3):S85-6.
- 12 Rocha V, Wagner JE, Sobocinski KA, *et al.* Graft-versus-host disease in children who have received a cord-blood or bone marrow transplant from an HLA-identical sibling. Eurocord and International Bone Marrow Transplant Registry working committee on alternative donor and stem cell sources. *N Engl J Med* 2000;342:1846-54.
- 13 Broxmeyer HE, Douglas GW, Hangoc G, *et al.* Human umbilical cord blood as a potential source of transplantable hematopoietic stem/progenitor cells. *Proc Natl Acad Sci U S A* 1989;86:3828-32.
- 14 Hows JM, Bradley BA, Marsh JC, *et al.* Growth of human umbilical-cord blood in longterm haemopoietic cultures. *Lancet* 1992;340:73-6.
- 15 Wang JC, Doedens M, Dick JE. Primitive human hematopoietic cells are enriched in cord blood compared with adult bone marrow or mobilized peripheral blood as measured by the quantitative in vivo SCID-repopulating cell assay. *Blood* 1997;89:3919-24.
- 16 Bhatia M, Wang JCY, Kapp U, *et al.* Purification of primitive human hematopoietic cells capable of repopulating immune-deficient mice. *Proc Natl Acad Sci U S A* 1997;94:5320-5.
- 17 Dick JE. Absence of CD34 on some human SCID-repopulating cells. *Ann N Y Acad Sci* 1999;872:211-7; discussion 217-9.
- 18 Wagner JE, Rosenthal J, Sweetman R, *et al.* Successful transplantation of HLA-matched and HLA-mismatched umbilical cord blood from unrelated donors: analysis of engraftment and acute graft-versus-host disease. *Blood* 1996;88:795-802.
- 19 Kurtzberg J, Laughlin M, Graham ML, *et al.* Placental blood as a source of hematopoietic stem cells for transplantation into unrelated recipients. *N Engl J Med* 1996;335:157-66.
- 20 Laughlin MJ, Rizzieri DA, Smith CA, *et al.* Hematologic engraftment and reconstitution of immune function post unrelated placental cord blood transplant in an adult with acute lymphocytic leukemia. *Leuk Res* 1998;22:215-19.
- 21 Cairo MS, Wagner JE. Placental and/or umbilical cord blood: an alternative source of hematopoietic stem cells for transplantation. *Blood* 1997;90:4665-78.
- 22 Donaldson C, Buchanan R, Webster J, *et al.* Development of a district cord blood bank: a model for cord blood banking in the National Health Service. *Bone Marrow Transplant* 2000;25:899-905.
- 23 Armitage S, Fehily D, Dickinson A, *et al.* Cord blood banking: volume reduction of cord blood units using a semi-automated closed system. *Bone Marrow Transplant* 1999;23:505-9.
- 24 Donaldson C, Armitage WJ, Denning-Kendall PA, *et al.* Optimal cryopreservation of human umbilical cord blood. *Bone Marrow Transplant* 1996;18:725-31.

- 25 Donaldson C, Armitage WJ, Laundry V, *et al.* Impact of obstetric factors on cord blood donation for transplantation. *Br J Haematol* 1999;106:128–32.
- 26 Bahceci E, Read EJ, Leitman S, *et al.* CD34+ cell dose predicts relapse and survival after T-cell-depleted HLA-identical haematopoietic stem cell transplantation (HSCT) for haematological malignancies. *Br J Haematol* 2000;108:408–14.
- 27 Denning-Kendall PA, Nicol A, Horsley H, *et al.* Is in vitro expansion of human cord blood cells clinically relevant? *Bone Marrow Transplant* 1998;21:225–32.
- 28 Donaldson C, Denning-Kendall P, Bradley B, *et al.* CD34+ expression overestimates CFU-GM content of cord blood donations expanded in serum-free conditions [abstract]. *Bone Marrow Transplant* 2000;25(suppl 1):A803.
- 29 Kutzberg J, Jaroscake J, Martin P, *et al.* Augmentation of umbilical cord blood (UCB) transplantation with ex-vivo expanded cells. A phase I trial using the replicell system [abstract]. *Blood* 1999;94(suppl 1):A2547.
- 30 Pecora AL, Stiff P, Jennis A, *et al.* Prompt and durable engraftment in two older adult patients with high risk chronic myelogenous leukemia (CML) using ex vivo expanded and unmanipulated unrelated umbilical cord blood. *Bone Marrow Transplant* 2000;25:797–9.
- 31 Lapidot T, Pflumio F, Doedens M, *et al.* Cytokine stimulation of multilineage hematopoiesis from immature human cells engrafted in SCID mice. *Science* 1992;255:1137–41.
- 32 Vormoor J, Lapidot T, Pflumio F, *et al.* SCID mice as an in vivo model of human cord blood hematopoiesis [see comments]. *Blood Cells* 1994;20:316–20; discussion 320–2.
- 33 Larochelle A, Vormoor J, Hanenberg H, *et al.* Identification of primitive human hematopoietic cells capable of repopulating NOD/SCID mouse bone marrow: implications for gene therapy. *Nat Med* 1996;2:1329–37.
- 34 Larochelle A, Vormoor J, Lapidot T, *et al.* Engraftment of immune-deficient mice with primitive hematopoietic cells from beta-thalassemia and sickle cell anemia patients: implications for evaluating human gene therapy protocols. *Hum Mol Genet* 1995;4:163–72.
- 35 Evelyn R, Chapman M, Denning-Kendall P, *et al.* Maintenance of SCID repopulating cell number following in-vitro expansion of cord blood [abstract]. *Br J Haematol* 1999;105(suppl 1):31.
- 36 Denning-Kendall P, Chapman M, Evelyn R, *et al.* Umbilical cord blood cells expanded with low dose cytokines in NOD/SCID mice [abstract]. *Haematology* 2000;1(suppl 1):A781.
- 37 van der Loo JC, Ploemacher RE. Marrow- and spleen-seeding efficiencies of all murine hematopoietic stem cell subsets are decreased by preincubation with hematopoietic growth factors. *Blood* 1995;85:2598–606.
- 38 Shen BJ. Human umbilical cord blood transplantation in 4 cases with advanced solid tumors. *Chung Hua Chung Liu Tsa Chih* 1993;15:152–4.
- 39 Weinreb S, Delgado JC, Clavijo OP, *et al.* Transplantation of unrelated cord blood cells. *Bone Marrow Transplant* 1998;22:193–6.
- 40 Armitage S, Warwick R, Fehily D, *et al.* Cord blood banking in London: the first 1000 collections. *Bone Marrow Transplant* 1999;24:139–45.
- 41 Aversa F, Tabilio A, Terenzi A, *et al.* Successful engraftment of T-cell-depleted haploidentical three-loci incompatible transplants in leukemia patients by addition of recombinant human granulocyte colony-stimulating factor-mobilized peripheral blood progenitor cells to bone marrow inoculum. *Blood* 1994;84:3948–55.

1st Asia Pacific Forum on Quality Improvement in Health Care

Three day conference

Wednesday 19 to Friday 21 September 2001

Sydney, Australia

We are delighted to announce this forthcoming conference in Sydney. Authors are invited to submit papers (call for papers closes on Friday 6 April), and delegate enquiries are welcome.

The themes of the Forum are:

- Improving patient safety
- Leadership for improvement
- Consumers driving change
- Building capacity for change: measurement, education and human resources
- The context: incentives and barriers for change
- Improving health systems
- The evidence and scientific basis for quality improvement.

Presented to you by the BMJ Publishing Group (London, UK) and Institute for Healthcare Improvement (Boston, USA), with the support of the the Commonwealth Department of Health and Aged Care (Australia), Safety and Quality Council (Australia), NSW Health (Australia), and Ministry of Health (New Zealand).

For more information contact: quality@bma.org.uk or fax +44 (0)20 7383 6869